AMENDMENT TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the applications:

Listing of Claims:

1-83. (canceled)

84. (currently amended) A method for assaying for modulators of β-secretase activity, comprising:

(a) contacting a polypeptide with β-secretase APP processing activity with a substrate, both in the presence and in the absence of a putative modulator compound;

wherein said substrate comprises an APP molecule having a modified β-secretase processing site defined by formula P_2P_1-$P_{1'}P_{2'}$, wherein:

P_2 ~~comprises an amino acid selected from the group consisting of~~ is N~~, L, K, S, G, T, D, A, Q and E~~;

P_1 ~~comprises an amino acid selected from the group consisting of Y, L, M, Nle,~~ is F ~~and H~~;

$P_{1'}$ ~~comprises an amino acid selected from the group consisting of E,~~ is A~~, D, M, Q, S and G~~;

$P_{2'}$ ~~comprises an amino acid selected from the group consisting of~~ is A~~, V, N, T, L, F and S~~;

wherein the substrate is cleaved between P_1 and $P_{1'}$ by a human aspartyl protease encoded by the nucleic acid sequence of SEQ ID NO: 1 or SEQ ID NO: 3 (Hu-Asp2); and

wherein said peptide does not comprise the corresponding P_2P_1-$P_{1'}P_{2'}$ portion of amino acid sequence depicted in SEQ ID NO: 19, SEQ ID NO: 20, SEQ ID NO: 21, SEQ ID NO: 26, SEQ ID NO: 27, SEQ ID NO: 28, SEQ ID NO: 31, SEQ ID NO: 32, SEQ ID NO: 33, SEQ ID NO: 34, SEQ ID NO: 35, SEQ ID NO: 36, SEQ ID NO: 37, SEQ ID NO: 38, or SEQ ID NO: 39;

(b) measuring cleavage of the substrate peptide in the presence and in the absence of the putative modulator compound; and

(c) identifying modulators of β-secretase activity from a difference in substrate cleavage in the presence versus in the absence of the putative modulator compound, wherein a modulator that is a β-secretase antagonist reduces such cleavage and a modulator that is a β-secretase agonist increases such cleavage.

85. (previously presented) The method of claim 84,

wherein the modified β-secretase processing site is defined by formula P_2P_1-$P_1'P_2'P_3'$, and

wherein P_3' comprises an amino acid selected from the group consisting of E, G, F, H, cysteic acid and S.

86. (canceled)

87. (currently amended) The method of claim [[84]] 85, wherein the modified β-secretase processing site is defined by the formula P_2P_1-$P_1'P_2'P_3'$, wherein

~~P_2 comprises an amino acid selected from the group consisting of S, N, and K;~~

~~P_1 comprises an amino acid selected from the group consisting of F, L, Y, and M;~~

~~P_1' comprises an amino acid selected from the group consisting of E, D and A;~~

~~P_2' comprises an amino acid selected from the group consisting of A and V;~~

P_3' is E.

88. (currently amended) The method of ~~any one of claims 84-87~~ claim 85, wherein the modified β-secretase processing site is defined by the formula $P_3P_2P_1$-$P_1'P_2'P_3'$, wherein P_3 is an amino acid selected from the group consisting of A, V, I, S, H, Y, T and F.

89. (previously presented) The method of claim 88, wherein P_3 comprises an amino acid selected from the group consisting of I or V.

90. (previously presented) The method of claim 88, wherein the modified β-secretase processing site is defined by the formula $P_4P_3P_2P_1$-$P_{1'}P_{2'}P_{3'}$ wherein P_4 is an amino acid selected from the group consisting of E, G, I, D, T, cysteic acid and S.

91. (previously presented) The method of claim 90, wherein the modified β-secretase processing site is defined by the formula $P_4P_3P_2P_1$-$P_{1'}P_{2'}P_{3'}P_{4'}$, wherein $P_{4'}$ is an amino acid selected from the group consisting of F, W, G, A, H, P, G, N, S, and E.

92 – 95. (canceled)

96. (currently amended) The method of any one of claims [[84-93]] 84, 85 or 87-91 wherein the APP molecule further comprises a first label.

97. (previously presented) The method of claim 96 wherein the APP molecule further comprises a second label.

98. (currently amended) The method of any one of claims [[84-93]] 84, 85 or 87-91 wherein the APP molecule further comprises a detectable label and a quenching moiety, wherein cleavage of the APP molecule between P_1 and $P_{1'}$ separates the quenching moiety from the label to permit detection of the label.

99. (previously presented) The method of claim 85, wherein said cysteic acid comprises a covalently attached label.

100. (currently amended) The method of any one of claims [[84-93]] 84, 85 or 87-91, wherein the rate of cleavage of said APP molecule by said human aspartyl protease is greater than the rate of cleavage of a polypeptide comprising the human APP β-secretase cleavage sequence: SEVKMDAEFR (SEQ ID NO: 20).

101. (currently amended) The method of any one of claims [[84-93]] 84, 85 or 87-91, wherein the rate of cleavage of said APP molecule by said human aspartyl protease is greater than the rate of cleavage of a polypeptide comprising the human APP Swedish KM→NL mutation, β-secretase cleavage sequence SEVNLDAEFR (SEQ ID NO: 19).

102. (currently amended) The method of any one of claims [[84-93]] 84, 85 or 87-91, wherein the polypepetide with β-secretase APP processing activity comprises an amino acid sequence selected from the group consisting of

(a) the amino acid sequence of SEQ ID NO: 2,

(b) a fragment of the amino acid sequence of SEQ ID NO: 2 that retains β-secretase APP processing activity, wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG,

(c) an amino acid sequence that is at least 95% identical to (a) or (b), wherein the polypeptide includes the aspartyl protease active site tripeptides DTG and DSG and exhibits β-secretase APP processing activity;

(d) the amino acid sequence SEQ ID NO: 4,

(e) a fragment of the amino acid sequence of SEQ ID NO: 4 that retains β-secretase APP processing activity, wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG, and

(f) an amino acid sequence that is at least 95% identical to (d) or (e), wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG and exhibits β-secretase APP processing activity.

103. (currently amended) The method of any one of claims [[84-93]] 84, 85 or 87-91, wherein the polypeptide with β-secretase APP processing activity comprises an amino acid sequence selected from the group consisting of

(a) the amino acid sequence of SEQ ID NO: 2; and

(b) a fragment of the amino acid sequence of SEQ ID NO: 2 that retains β-secretase APP processing activity, wherein said fragment includes the aspartyl protease active site tripeptides DTG and DSG.

104. (previously presented) A method according to claim 103, wherein the polypeptide with β-secretase APP processing activity comprises a polypeptide purified and isolated from a cell transformed or transfected with a polynucleotide comprising a nucleotide sequence that encodes the polypeptide.

105. (previously presented) A method according to claim 95,

wherein the APP molecule is expressed in a cell transformed or transfected with a polynucleotide comprising a nucleotide sequence that encodes the APP molecule,

wherein the cell expresses the polypeptide with β-secretase APP processing activity;

wherein the contacting comprises growing the cell in the presence and absence of the test agent, and

wherein the measuring step comprises measuring APP processing activity of the cell.

106. (previously presented) A method according to claim 105, wherein the contacting comprises administering the test agent to a transgenic non-human mammal that comprises the cell.

107. (previously presented) A method according to claim 84, wherein the polypeptide is encoded by a polynucleotide comprising the nucleotide sequence selected from the group consisting of:

(a) the nucleotide sequence of SEQ ID NO: 1 or SEQ ID NO; 3,

(b) a nucleotide sequence that hybridizes under the following stringent hybridization conditions to the complement of SEQ ID NO: 1 or 3:

(1) hybridization at 42°C in a hybridization buffer comprising 6x SSC and 0.1% SDS, and

(2) washing at 65°C in a wash solution comprising 1x SSC and 0.1% SDS;

wherein said nucleotide sequence encodes a polypeptide that exhibits β-secretase APP processing activity.

108. (previously presented) The method of claim 84, wherein the modified β-secretase processing site is defined by the formula $P_4P_3P_2P_1$-$P_{1'}P_{2'}P_{3'}P_{4'}$ as provided by SEQ ID NO: 5, SEQ ID NO: 7, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48, SEQ ID NO: 115, SEQ ID NO: 116, SEQ ID NO: 117, SEQ ID NO: 118, SEQ ID NO: 119, SEQ ID NO: 133, SEQ ID NO: 135, SEQ ID NO: 136, SEQ ID NO: 137, SEQ ID NO: 141, SEQ ID NO: 143, SEQ ID NO: 144, SEQ ID NO: 145, SEQ ID NO: 147, SEQ ID NO: 149, SEQ ID NO: 150, SEQ ID NO: 151, SEQ ID NO: 152 or SEQ ID NO: 153.

109. (canceled)

110. (previously amended) The method of claim 88, wherein the peptide comprises a sequence of amino acids defined by the formula $P_3P_2P_1$-$P_{1'}P_{2'}P_{3'}$, wherein P_3 is V, P_2 is N, P_1 is F, $P_{1'}$ is A, $P_{2'}$ is A and $P_{3'}$ is E.